EAST Search History

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	2	"6656726".pn.	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2006/05/15 11:01
L2	43	126/183	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2006/05/15 11:01
L3	32	126/183 and viral	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2006/05/15 11:01
L4	32	126/183 and viral and plant	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2006/05/15 11:01

US005977438A

United States Patent [19]

Turpen et al.

[11] Patent Number: 5,977,438

[45] Date of Patent: Nov. 2, 1999

[54] **PRODUCTION OF PEPTIDES IN PLANTS AS VIRAL COAT PROTEIN FUSIONS**

[75] Inventors: **Thomas H. Turpen**, Vacaville; **Stephen J. Reinl**, Sacramento; **Laurence K. Grill**, Vacaville, all of Calif.

[73] Assignee: **Biosource Technologies, Inc.**, Vacaville, Calif.

[21] Appl. No.: **08/324,003**

[22] Filed: **Oct. 14, 1994**

Related U.S. Application Data

[63] Continuation-in-part of application No. 08/176,414, Dec. 29, 1993, Pat. No. 5,811,653, and application No. 08/184,237, Jan. 19, 1994, Pat. No. 5,589,367, said application No. 08/176,414, is a continuation-in-part of application No. 07/997,733, Dec. 30, 1992, abandoned, said application No. 08/184,237, is a continuation-in-part of application No. 07/997,733, Dec. 30, 1992, abandoned, which is a continuation of application No. 07/923,692, Jul. 31, 1992, Pat. No. 5,316,931, which is a continuation-in-part of application No. 07/600,244, Oct. 22, 1990, abandoned, application No. 07/641,617, Jan. 16, 1991, abandoned, application No. 07/737,899, Jul. 26, 1991, abandoned, and application No. 07/739,143, Aug. 1, 1991, abandoned, said application No. 07/600,244, is a continuation of application No. 07/310,881, Feb. 17, 1989, abandoned, which is a continuation-in-part of application No. 07/160,766, Feb. 26, 1988, abandoned, and application No. 07/160,771, Feb. 26, 1988, abandoned, said application No. 07/641,617, is a continuation of application No. 07/347,637, May 5, 1989, abandoned, said application No. 07/737,899, is a continuation of application No. 07/363,138, Jun. 8, 1989, abandoned, which is a continuation-in-part of application No. 07/219,279, Jul. 15, 1988, abandoned, said application No. 07/739,143, is a continuation-in-part of application No. 07/600,244, Oct. 22, 1990, abandoned, application No. 07/641,617, Jan. 16, 1991, abandoned, and application No. 07/737,899, Jul. 26, 1991, abandoned.

[51] **Int. Cl.**[6] **A01H 5/00**; C12N 5/04; C12N 15/40; C12N 15/62; C12N 15/83

[52] **U.S. Cl.** **800/288**; 800/278; 800/298; 536/23.4; 536/23.5; 536/23.72; 435/69.7; 435/70.1; 435/235.1; 435/419; 435/468

[58] **Field of Search** 536/23.4, 23.5, 536/23.72, 24.1; 435/69.1, 70.1, 172.3, 235.1, 240.4, 418, 419, 69.7, 468; 800/205, 278, 288, 298

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(List continued on next page.)

Primary Examiner—David T. Fox

Attorney, Agent, or Firm—Albert P. Halluin; John A. Bendrick; Howrey & Simon

[57] **ABSTRACT**

The present invention relates to foreign peptide sequences fused to recombinant plant viral structural proteins and a method of their production. Fusion proteins are economically synthesized in plants at high levels by biologically contained tobamoviruses. The fusion proteins of the invention have many uses. Such uses include use as antigens for inducing the production of antibodies having desired binding properties, e.g., protective antibodies, or for use as vaccine antigens for the induction of protective immunity, including immunity against parasitic infections.

19 Claims, 10 Drawing Sheets

-continued

```
Leu Ile Val Glu Leu Ile Arg Gly Thr Gly Ser Tyr Asn Arg Ser Ser
    130                 135                 140

Phe Glu Ser Ser Ser Gly Leu Val Trp Thr Ser
145                 150                 155
```

What is claimed is:

1. A polynucleotide encoding a fusion protein capable of being expressed in a plane or a plant cell, wherein the fusion protein comprises a plant viral coat protein from a single-stranded plus-sense RNA virus fused to a protein of interest comprising four or more amino acids, said polynucleotide further comprising a promoter functional in plants 5' to the fusion protein encoding region.

2. A polynucleotide according to claim 1, encoding a fusion protein wherein the protein of interest is amino-terminal to the plant viral coat protein.

3. A polynucleotide according to claim 1, encoding a fusion protein wherein the protein of interest is carboxy-terminal to the plant viral coat protein.

4. A polynucleotide according to claim 1, wherein said fusion protein is an internal fusion protein.

5. A polynucleotide according to claim 1, further comprising a fusion joint having a leaky stop codon from a single-stranded pulse-sense RNA virus.

6. A polynucleotide according to claim 1, wherein the protein of interest is an antigen.

7. A polynucleotide according to claim 1, wherein the coat protein is a tobacco mosaic virus coat protein.

8. A recombinant plant viral genome comprising a polynucleotide according to claim 1.

9. A recombinant plant virus particle, comprising a genome according to claim 8.

10. A recombinant plant virus, wherein the coat protein is encoded by a polynucleotide according to claim 1.

11. A plant cell comprising a polynucleotide according to claim 8.

12. A polynucleotide according to claim 1 wherein the coat protein is a tobamovirus coat protein.

13. A plant cell comprising a recombinant plant viral genome according to claim 8.

14. A plant cell comprising a recombinant plant virus particle according to claim 9.

15. A plant cell comprising a recombinant plant virus according to claim 10.

16. A plant comprising a polynucleotide according to claim 1.

17. A plant comprising a recombinant plant viral genome according to claim 8.

18. A plant comprising a recombinant plant virus particle according to claim 9.

19. A plant comprising a recombinant plant virus according to claim 10.

* * * * *

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT NO. : 5,977,438
DATED : November 2, 1999
INVENTOR(S) : Thomas H. Turpen, et al.

It is certified that error appears in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

Column 2:
Line 43, change "plan" to --plant--;

Column 7:
Line 15, change "*alciparum*" to --*falciparum*--;
Line 18, change"accines" to --vaccines--;

Column 35:
Line 11, change "plane" to --plant--; and
Line 27, change "pluse-sense" to --plus-sense--.

Signed and Sealed this

Nineteenth Day of June, 2001

Attest:

Nicholas P. Godici

Attesting Officer

NICHOLAS P. GODICI
Acting Director of the United States Patent and Trademark Office

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Parent Data

10624193
is a continuation of 09565616
Which Claims Priority from Provisional Application 60132697

Child Data

No Child Data

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Inventor Information for 10/624193

Inventor Name	City	State/Country
FITZMAURICE, WAYNE P.	VACAVILLE	CALIFORNIA
POGUE, GREGORY P.	VACAVILLE	CALIFORNIA
LINDBO, JOHN A.	VACAVILLE	CALIFORNIA

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Inventor Name Search Result

Your Search was:

Last Name = LINDBO
First Name = JOHN

Application#	Patent#	Status	Date Filed	Title	Inventor Name
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US006656726B1

(12) **United States Patent**
Fitzmaurice et al.

(10) **Patent No.: US 6,656,726 B1**
(45) **Date of Patent: Dec. 2, 2003**

(54) **VIRAL EXPRESSION VECTORS**

(75) Inventors: **Wayne P. Fitzmaurice**, Vacaville, CA (US); **Gregory P. Pogue**, Vacaville, CA (US); **John A. Lindbo**, Vacaville, CA (US)

(73) Assignee: **Large Scale Biology Corporation**, Vacaville, CA (US)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 0 days.

(21) Appl. No.: **09/565,616**

(22) Filed: **May 4, 2000**

Related U.S. Application Data

(60) Provisional application No. 60/132,697, filed on May 4, 1999.

(51) **Int. Cl.**[7] **C12N 15/00**; C12N 5/00; C12N 5/04; C12N 15/82

(52) **U.S. Cl.** **435/320.1**; 435/410; 435/419; 435/468

(58) **Field of Search** 536/23.2, 23.72; 800/295, 278, 298, 317.3; 435/419, 468, 410, 414, 320.1

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Primary Examiner—Deborah Crouch
Assistant Examiner—Joseph Woitach
(74) *Attorney, Agent, or Firm*—John C. Robbins; Thomas Gallegos; Quine Intellectual Property Law Group, P.C.

(57) **ABSTRACT**

The present invention provides nucleic acid sequences having an altered viral movement protein and 126/183 kDa replicase proteins further characterized in its ability to stabilize a transgene contained in a virus that expresses the altered movement protein. The present invention also provides viral vectors expressing the altered movement protein, cells transformed with the vectors, and host plants infected by the viral vectors.

14 Claims, 17 Drawing Sheets

-continued

```
Asn Ser Ser Ser Asp Arg Ser Val Pro Asn Lys Asn Tyr Arg Asn Val
225                 230                 235                 240

Lys Asp Phe Gly Gly Met Ser Phe Lys Lys Asn Asn Leu Ile Asp Asp
                245                 250                 255

Asp Ser Glu Ala Thr Val Ala Glu Ser Asp Ser Phe
            260                 265


<210> SEQ ID NO 6
<211> LENGTH: 268
<212> TYPE: PRT
<213> ORGANISM: Nicotiana tabacum

<400> SEQUENCE: 6

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 1               5                  10                  15

Leu Thr Lys Met Glu Lys Ile Leu Pro Ser Met Glu Thr Pro Val Lys
            20                  25                  30

Ser Val Met Cys Ser Lys Val Asp Lys Ile Met Val His Glu Asn Glu
        35                  40                  45

Ser Leu Ser Gly Val Asn Leu Leu Lys Gly Val Lys Leu Ile Asp Ser
    50                  55                  60

Gly Tyr Val Cys Leu Ala Gly Leu Val Val Thr Gly Glu Trp Asn Leu
65                  70                  75                  80

Pro Asp Asn Cys Arg Gly Gly Val Ser Val Cys Leu Val Asp Lys Arg
                85                  90                  95

Met Glu Arg Ala Asp Glu Ala Ile Leu Gly Ser Tyr Tyr Thr Ala Ala
            100                 105                 110

Ala Lys Lys Arg Phe Gln Phe Lys Val Val Pro Asn Tyr Ala Ile Thr
        115                 120                 125

Thr Gln Asp Ala Met Arg Asn Val Trp Gln Val Leu Val Asn Ile Arg
    130                 135                 140

Asn Val Lys Met Ser Ala Gly Phe Cys Pro Leu Ser Leu Glu Phe Val
145                 150                 155                 160

Ser Val Cys Ile Val Tyr Arg Asn Asn Ile Lys Leu Gly Leu Arg Glu
                165                 170                 175

Lys Ile Thr Asn Val Arg Asp Gly Gly Pro Met Glu Leu Thr Glu Glu
            180                 185                 190

Val Val Asp Glu Phe Met Glu Asp Val Pro Met Ser Ile Arg Leu Ala
        195                 200                 205

Lys Phe Arg Ser Arg Thr Gly Lys Lys Ser Asp Val Arg Lys Gly Lys
    210                 215                 220

Asn Ser Ser Ser Asp Arg Ser Val Pro Asn Lys Asn Tyr Arg Asn Val
225                 230                 235                 240

Lys Asp Phe Gly Gly Met Ser Phe Lys Lys Asn Asn Leu Ile Asp Asp
                245                 250                 255

Asp Ser Glu Ala Thr Val Ala Glu Ser Asp Ser Phe
            260                 265
```

What is claimed is:

1. An isolated nucleic acid sequence comprising a nucleic acid sequence encoding an altered viral movement protein having the amino acid sequence shown in SEQ ID NO: 6.

2. The isolated nucleic acid sequence of claim 1 that is identical to the sequence shown in SEQ ID NO: 4.

3. The isolated nucleic acid of claim 1, wherein the altered movement protein enhances the ability to facilitate stabilization of a transgene contained in a virus that expresses the altered movement protein.

4. An isolated nucleic acid sequence comprising a nucleic acid sequence encoding an altered 126/183 replicase complex having a nucleic acid alteration at nucleotide positions 1138, 1268, 2382, and 3632 as shown in SEQ ID NO: 2 .

5. An isolated nucleic acid according to claim 4 wherein the altered 126/183 replicase complex enhances the stabili-.

zation of a transgene contained in a virus that expresses the altered replicase complex.

6. A viral vector comprising a nucleic acid sequence encoding an altered viral movement protein having the amino acid sequence shown in SEQ ID NO.: 6.

7. The viral vector of claim 6, further comprising a transgene, wherein said viral vector exhibits an enhanced ability to stabilize said transgene compared to a control viral vector comprising a wild type movement protein as shown in SEQ ID NO: 3.

8. The viral vector of claim 7, wherein the transgene is a non-viral gene.

9. The viral vector of claim 8, wherein the non-viral transgene encodes a protein selected from the group consisting of a membrane protein, a cytosolic protein, a secreted protein, a nuclear protein, and a chaperon protein.

10. The viral vector of claim 6, wherein the vector is a tobacco mosaic viral vector.

11. The viral vector of claim 6 that is designated BSG1057 deposited with American Type Culture Collection accession number 20398.

12. A plant cell transformed with the viral vector of claim 6.

13. An isolated nucleic acid sequence of SEQ ID NO: 1.

14. An isolated nucleic acid sequence of SEQ ID NO: 2.

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